Ecography

ECOG-03975
Fitt, R. N. L., Palmer, S., Hand, C., Travis, J. M. J. and Lancaster, L. T. 2019. Towards an interactive, process-based approach to understanding range shifts: developmental and environmental dependencies matter. – Ecography doi: 10.1111/ecog.03975

Supplementary material

Appendix 1

Supplemental methods and results associated with the motivational study (Box 1):

Collection and rearing protocols

We captured wild Ischnura elegans (Van der Linden 1820) damselfly larvae from three sites in northeast Scotland, Midmar (Lat: 57° 6’ 27”; Long: 20° 30’ 24”, n=97), Raemoir (Lat: 57° 4’ 9”; Long: 20° 30’ 12”, n=26) and Mossat (Lat: 57° 15’ 54”; Long: 20° 52’ 28”,n=100) while the larvae were still in a diapause state (daytime water temperature at collection time was 4°C, below the temperature at which larvae are expected to emerge from diapause; Thompson, 1978). At capture, individuals were identified to *Ischnura elgans,* although an additional 168 were mis-identified *Enallagma cyathigerum.* This error was discovered at the stage of adult emergence, and thus mis-identified individuals were removed from the data where possible. Body size was estimated via head width measurements using a Yenway SZN 71 microscope and YenCam CMOS mounted camera. Head width was used as a measure of body size, as it is easily measured accurately and is strongly proportional to body length (Thompson 1978). Larvae were then assigned to cohorts based on head width at capture (see statistical methods and results). Larvae were placed in individual, ventilated containers with 100ml of dechlorinated tap water (tap water which had been left to stand for at least 48 hours) and kept in an LMS series 3 model 300 incubator at 4°C and a photoperiod of 8 hours light & 16 hours dark, mimicking field conditions to maintain diapause. Larvae were randomly assigned to one of two diapause treatments, short (i.e., maintained in diapause conditions for 33 days post capture) and long (68 days post capture), after which time the larvae were removed from diapause by increasing the temperature up to 20°C, which is relatively high for Scottish freshwater systems in Spring, but reflects a sensible temperature under a 4°C warming scenario (Hrachowitz et al. 2010), and photoperiod to 12L:12D, with temperature increasing by 2.29°C per day and daylight increasing by 34.2 minutes per day over seven days (Figure 1). Due to the need to conduct separate temperature ramping treatments for each treatment, the two treatments (short- and long-diapause) were housed in separate but identical incubators. The temperature of each incubator was monitored with a thermal data logger (HOBO Onset UA-001-64)

to ensure identical thermal treatments. Weekly, 20% water changes were performed with dechlorinated tap water.

Post diapause, larvae were fed daily ad libitum on a rotating diet of: *Aedes aegypti* larvae, Copepoda spp, and *Artemia* spp. Head width was measured weekly until the damselflies were ready to emerge or had died, at which point the date was recorded. Damselflies were assessed to be ready to emerge as adults when they were in their final instar, identified by fully developed wing buds, and refusal of food. When this occurred, the lid was removed from the damselfly's container, and the container was placed in a 10 cm (diameter) x 20 cm (height) cylindrical mesh cage with a perch inserted for the individual to climb during emergence. The container was then placed back into the incubator, and emerging damselflies were checked daily. Once emerged, each damselfly was allowed 24 hours at room temperature for its wings to harden before further study.

Adult flight performance and size

Once each damselfly had emerged and its wings hardened, it was placed in a 10 cm (diameter) x 20 cm (height) cage in a controlled environment room with the temperature set for optimum flight performance (27°C; Taylor 1963), and allowed to equilibrate for 30 minutes. Once equilibrated, flight endurance behaviour was measured using methods described by Ducatez et al. (2013), whereby each damselfly was placed in a 10 cm (diameter) X 30 cm (height) cylinder, and this cylinder was agitated using a vortex mixer on half power, forcing the damselfly to fly. The time was then recorded from the point of its taking flight to the point it refused to fly any further. This endurance trial was replicated for each damselfly three times with a 5 minute recovery time between trials. All adult damselflies survived the experimental procedure.

Morphological measurements of adults were performed following behavioural trials, by scanning the dorsal view of each temporarily-immobilised damselfly using an Epson Perfection V37 flatbed scanner, at 600 DPI resolution. From these scans, measurements were taken of forewing length, abdomen length and width, thorax width, and total body length using ImageJ (Schneider et al. 2012). Principal component analysis of these measurements was conducted in R 3.2.3 (R Development Core Team 2012 to summarise variation in body size and shape (Gosden et al., 2011). The first principal component (PC1), accounting for 50.3% of the total variance and positively correlated with each morphological measurement, was used as an index of body size. The second principal component (PC2) accounted for 22.6% of the total variance and mapped to differences in body shape (higher values of PC2 had longer/leaner body shapes).

Statistical analysis

Individuals were classified via visual inspection of a histogram of size (Figure 1A) into cohorts based on larval size at collection, with individual body sizes assigned to one of three discrete distributions of body size within each cohort. Drivers of larval post-diapause survival and adult body size were analysed using a linear mixed model incorporating fixed effects of cohort, diapause treatment, and sex, and a random effect for collection site. Drivers of flight endurance time were similarly assessed using a linear mixed model, including fixed effects of diapause treatment, sex, size at collection and adult size, and random intercepts for individual and collection site. Analyses were performed using the lme4 (Bates et al., 2015) and lmerTest (Kuznetsova et al., 2016) packages for R v.3.2.3 (R Development Core Team 2012).

Figure A1. Larval developmental experimental design schematic

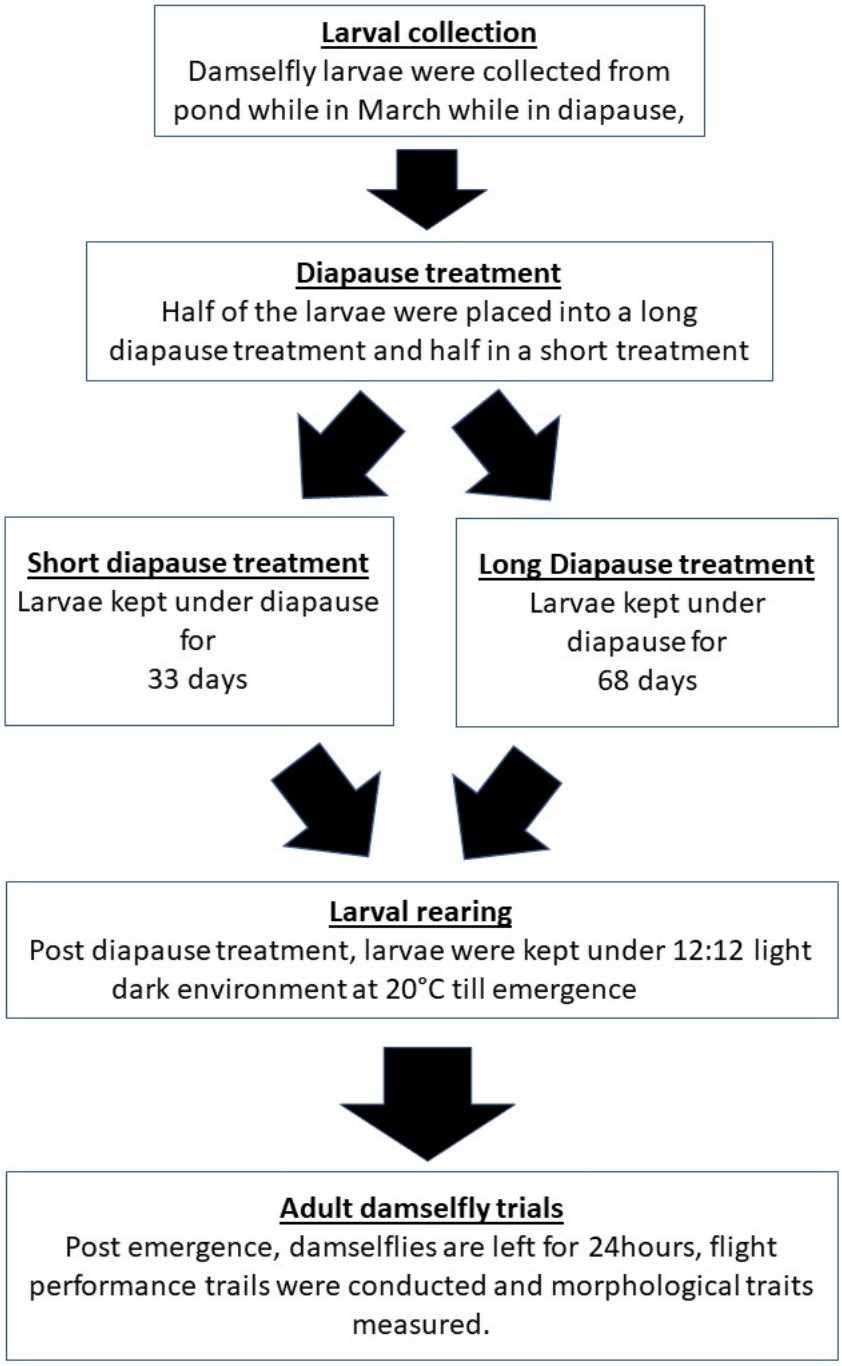

Table A1 Simulated values of fecundity, mean of the dispersal kernel, and emigration probability used in the model. The values "Low" and "High" at the bottom of column 1 refer to changes in trait values as a result of differing developmental sensitivities.

	Fecundity	Mean of the dispersal Kernel	Emigration probability
		Developmental strategy (Years)	

Env	1	2	3	1	2	3	1	2	3
1-stressful	4.5	8.5	12.5	150	290	430	0.088	0.188	0.288
2	5	9	13	170	310	450	0.104	0.204	0.304
3	5.5	9.5	13.5	190	330	470	0.12	0.22	0.32
4	6	10	14	210	350	490	0.136	0.236	0.336
5	6.5	10.5	14.5	230	370	510	0.152	0.252	0.352
6	7	11	15	250	390	530	0.168	0.268	0.368
7	7.5	11.5	15.5	270	410	550	0.184	0.284	0.384
8	8	12	16	290	430	570	0.2	0.3	0.4
9	8.5	12.5	16.5	310	450	590	0.216	0.316	0.416
10-benign	9	13	17	330	470	610	0.232	0.332	0.432
Low	+2	0	-2	+50	0	+50	+0.06	0	-0.06
High	-2	0	+2	-50	0	-50	-0.06	0	+0.06

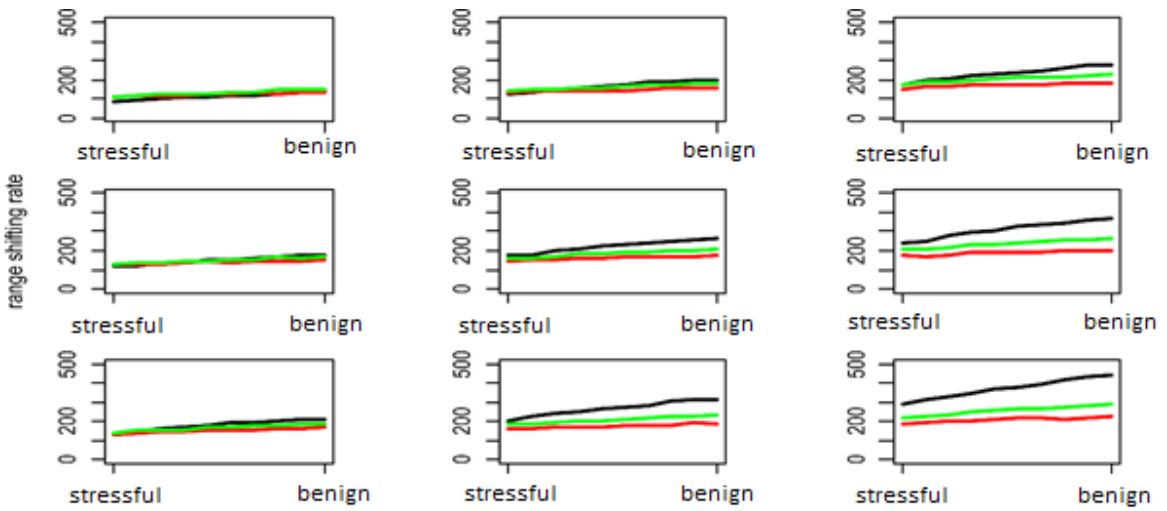

Figure A2. Modelled rates of range expansion that would be obtained by populations exhibiting each of the three developmental strategies and living in a particular environmental condition, when fecundity is allowed to vary over the environmental gradient, and emigration probability is either low (top left), medium or high (top right) and dispersal distance either low (bottom left) , medium or high (bottom right). Green line = 1-year developmental strategy, blue line = 2-year developmental strategy, red line = 3-year strategy.

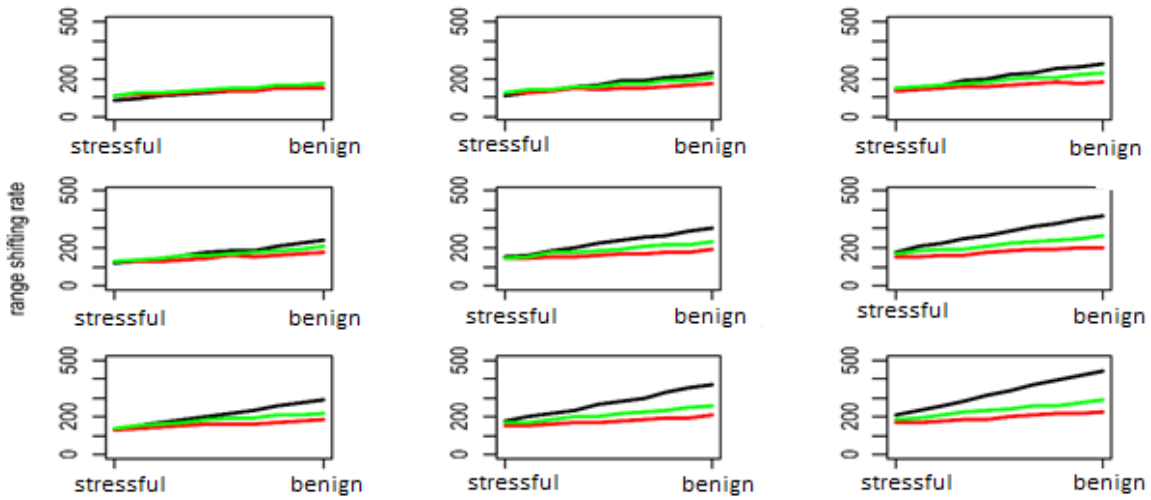

Figure A3. Modelled rates of range expansion that would be obtained by populations exhibiting each of the three developmental strategies and living in a particular environmental condition, when dispersal distance is allowed to vary over the environmental gradient, and emigration probability is either low, medium or high (left to right) and fecundity is either low, medium or high (top to bottom). Green line = 1-year developmental strategy, blue line = 2-year developmental strategy, red line = 3-year strategy.

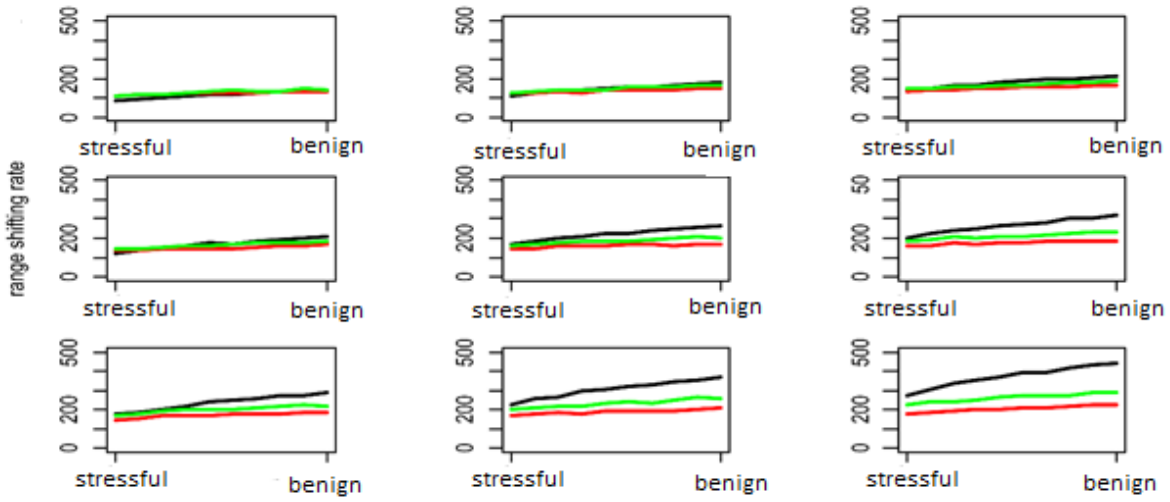

Figure A4. Modelled rates of range expansion that would be obtained by populations exhibiting each of the three developmental strategies and living in a particular environmental condition, when emigration probability is allowed to vary over the environmental gradient, and dispersal distance is either low, medium or high (left to right) and fecundity is either low, medium or high (top to bottom). Green line = 1-year developmental strategy, blue line = 2-year developmental strategy, red line = 3-year strategy.

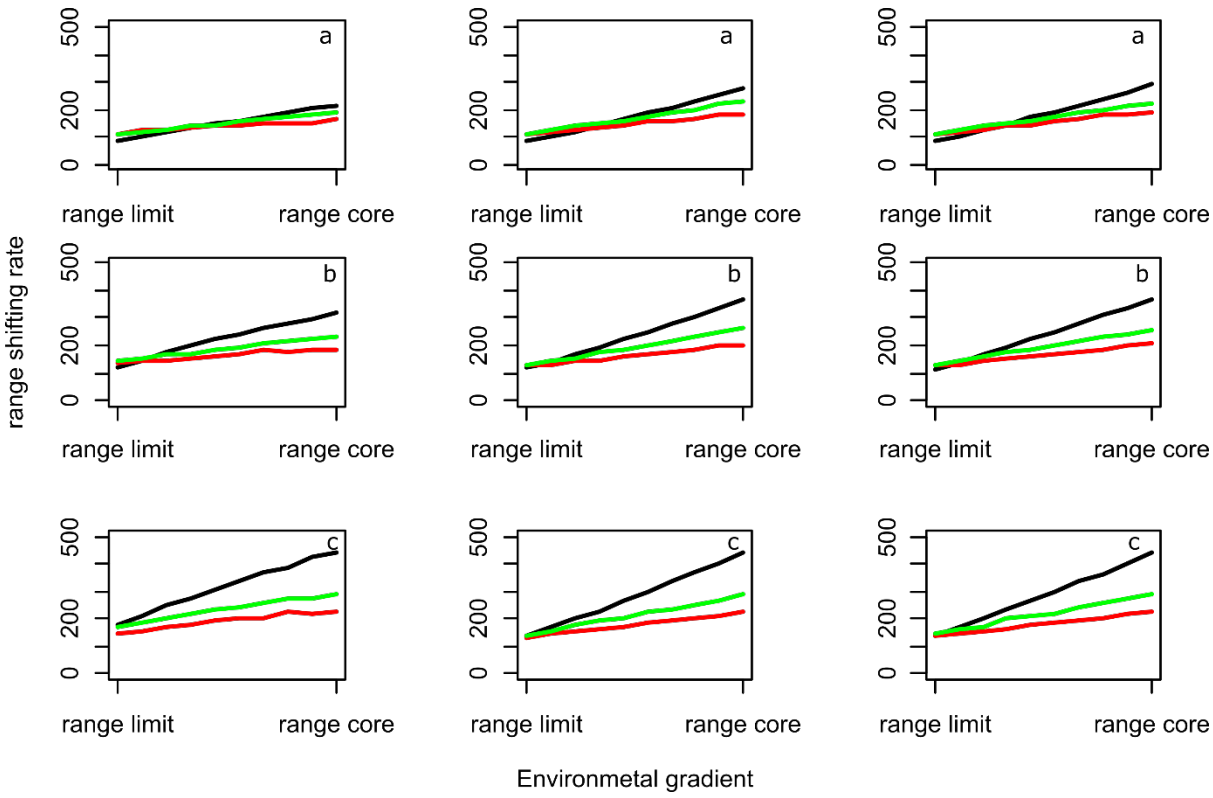

Figure A5. Modelled rates of range expansion that would be obtained by populations exhibiting each of the three developmental strategies and living in a particular environmental condition, when a) emigration probability and mean of the dispersal kernel are allowed to vary, and fecundity is either low medium or high (left to right) b) emigration probability and fecundity are allowed to vary, and dispersal distance is either low, medium or high c) mean of the dispersal kernel and fecundity are allowed to vary over the environmental gradient and emigration probability is either low, medium or high. Green line = 1-year developmental strategy, blue line = 2-year developmental strategy, red line = 3-year strategy.

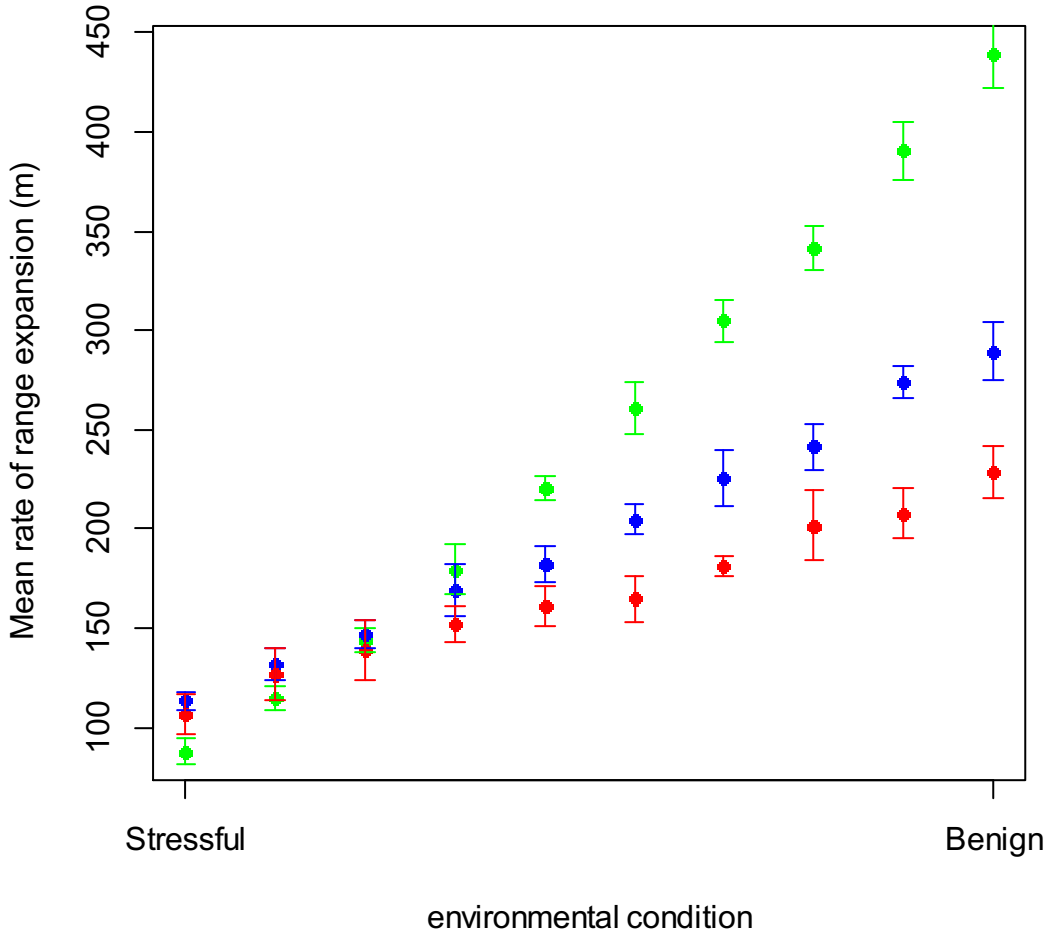

Figure A6. Modelled rates of range expansion under moderate developmental dependence and environmental dependence of all traits, where green points represents a one year developmental strategy, blue points represents a two year developmental strategy and red points represents a three year strategy. Error bars depict Standard deviation.